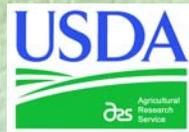


# **Comparison of Batch and Flow Experimental Data on Retention of Manure-borne *Cryptosporidium parvum* oocysts in soils**

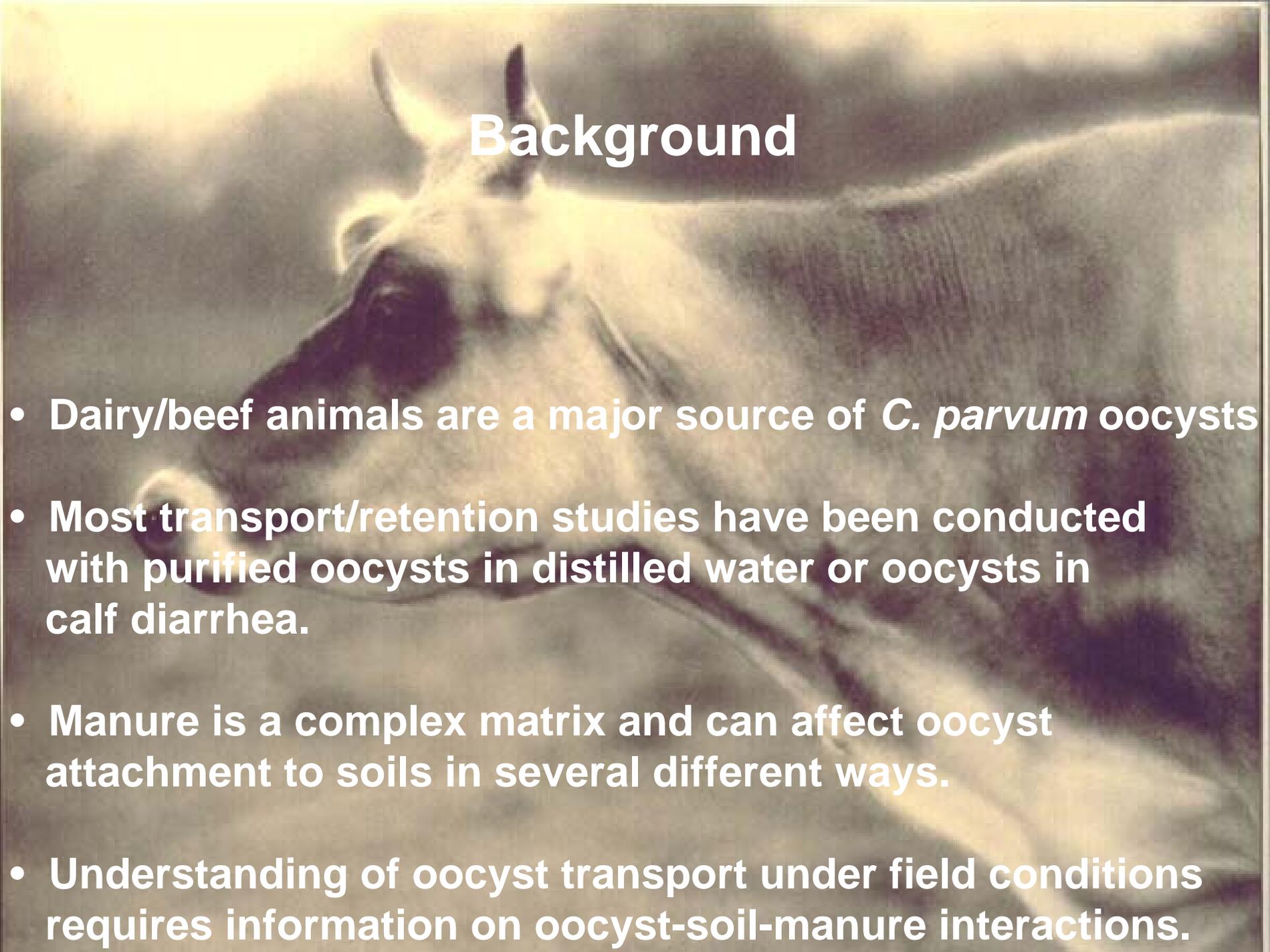
**Yakov Pachepsky and Daniel Shelton**

**USDA-Agricultural Research Service  
Environmental Microbial Safety Laboratory, Beltsville, MD**



# LAYOUT

- 1. Background**
- 2. Batch experiments with two soils**
- 3. Column experiments**  
**with saturated and unsaturated soils**
- 4. Parameter estimation and comparison**
- 5. Conclusions**



## Background

- Dairy/beef animals are a major source of *C. parvum* oocysts
- Most transport/retention studies have been conducted with purified oocysts in distilled water or oocysts in calf diarrhea.
- Manure is a complex matrix and can affect oocyst attachment to soils in several different ways.
- Understanding of oocyst transport under field conditions requires information on oocyst-soil-manure interactions.

# Batch experiments

- 1. Dairy manure was diluted to obtain 1% and 10% suspensions.**
- 2. Manure suspensions were inoculated with purified *C. parvum* oocysts to have concentrations ca.  $10^5$  oocysts/mL.**
- 3. Nine ml of sandy loam and clay loam soil 1% suspensions were mixed 1 ml with manure suspensions, vortexed thoroughly, and incubated for 2 h with a gentle shaking.**
- 4. Suspensions were centrifuged for 10 min at 100 g, the top 9 mL of supernatant was removed using a glass pipette and 9 ml of distilled water added to tubes. The Step 3 above was repeated.**
- 5. Oocysts were enumerated as previously described by Kuczynska and Shelton (1999).**

**Two replications were conducted independently with different bovine manures from Beltsville, MD.**

**Data inspection showed that the data for two manures are essentially identical.**

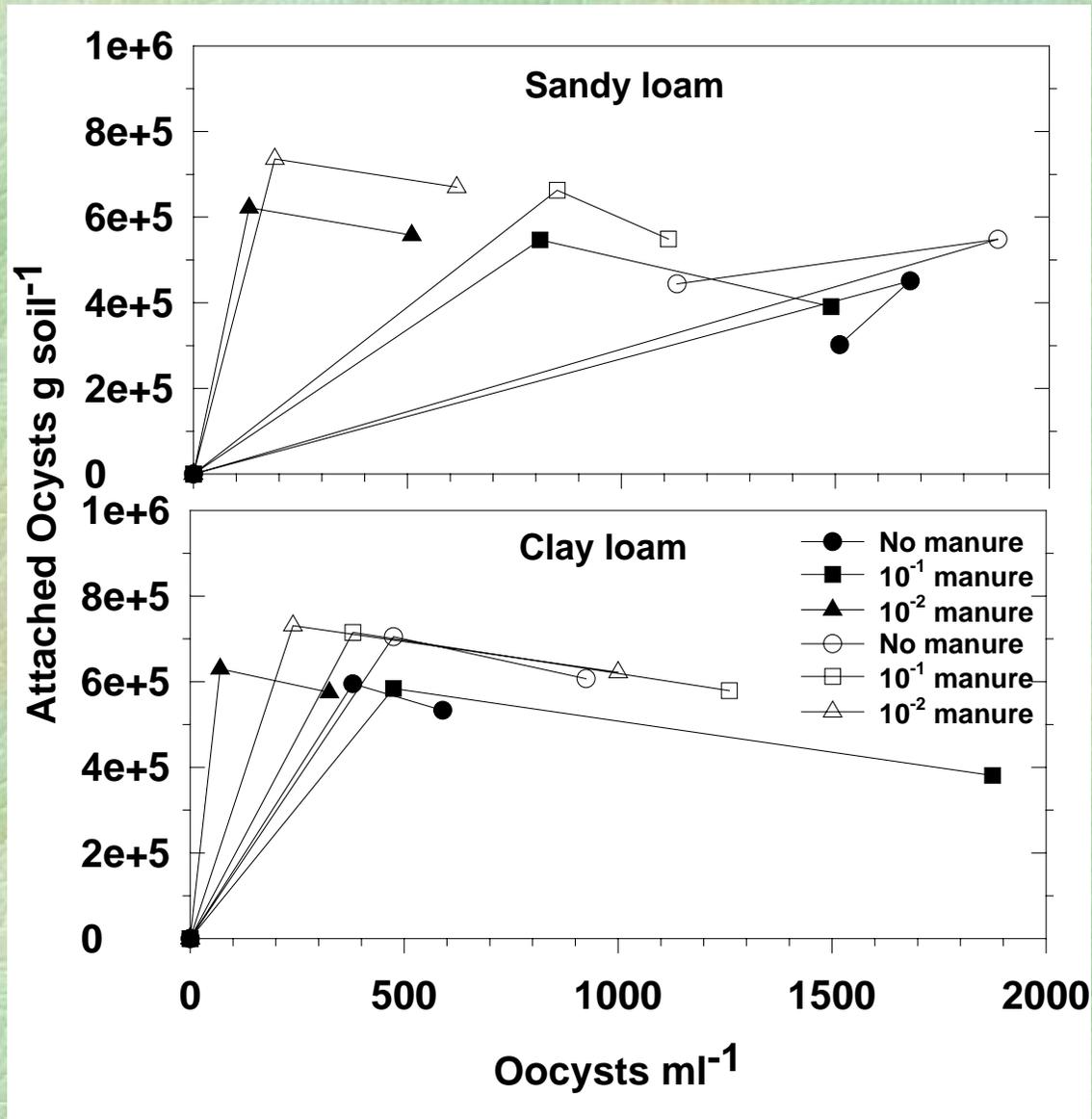
**ANOVA supported this conclusion; the manure concentration and the soil texture were significant factors at  $P < 0.001$**

# Percentage of oocysts attached to soil particles

<u>Treatment</u>	<u>Sandy Loam (<math>K_d</math>)</u>	<u>Clay Loam (<math>K_d</math>)</u>
No manure	72.0±3.6 <sup>1</sup> (280)	93.1±2.9 (1520)
1.0% manure	86.7±2.2 (730)	93.1±3.8 (1360)
0.1% manure	97.4±1.8 (4300)	97.7±2.0 (6000)

<sup>1</sup>Mean and S.D.

# Oocyst attachment to soil particles in presence and in absence of manure



# Column experiments

Columns 10 cm long and 11.5 cm in diameter were packed with air-dried and sieved clay loam and sandy loam soil to 50% porosity.

Columns were slowly saturated from the bottom.

50 grams of bovine manure, seeded with  $7.1 \times 10^6$  oocysts, were applied to the surface of soil columns and covered with straw.

Rainfall was applied at ca. 1.6 cm/h for 8 hours.

Saturated flow and unsaturated flow at 5 kPa suction were created for each soil.

Leachate was collected at one-hour intervals.

At the end of the experiment, the manure on the surface of cores was collected and cores were sliced into five 2 cm sections.

Leachate and soil samples were processed as previously described (Kuczynska and Shelton, 1999).

# Data analysis

$$\frac{\partial}{\partial t}(\theta_a c + \rho s) = \frac{\partial}{\partial x} \left( \theta_a D \frac{\partial c}{\partial x} - J_w c \right) - \mu_l \theta_a c - \rho \mu_s s$$

$$s = K_d c$$

---

$$R \frac{\partial c}{\partial t} = \frac{\theta_a}{\theta} D \frac{\partial^2 c}{\partial x^2} - v \frac{\partial c}{\partial x} - \mu c$$

$$R = \frac{\theta_a}{\theta} + \frac{\rho K_d}{\theta}$$

$$\mu = \mu_l \frac{\theta_a}{\theta} + \frac{\rho K_d \mu_s}{\theta}$$

---

$$c_m(t) = c_0 \exp(-\lambda t)$$

$\theta_a$  = the porosity available for oocyst transport

$\rho$  = soil bulk density

$s$  = the adsorbed amount of oocyst,

$D$  = the dispersion coefficient

$J_w$  = the volumetric water flux density

$\mu_l$  and  $\mu_s$  = first-order removal rate constants

$K_d$  = the distribution constant

$v$  = the average pore-water velocity

$R$  = the retardation factor

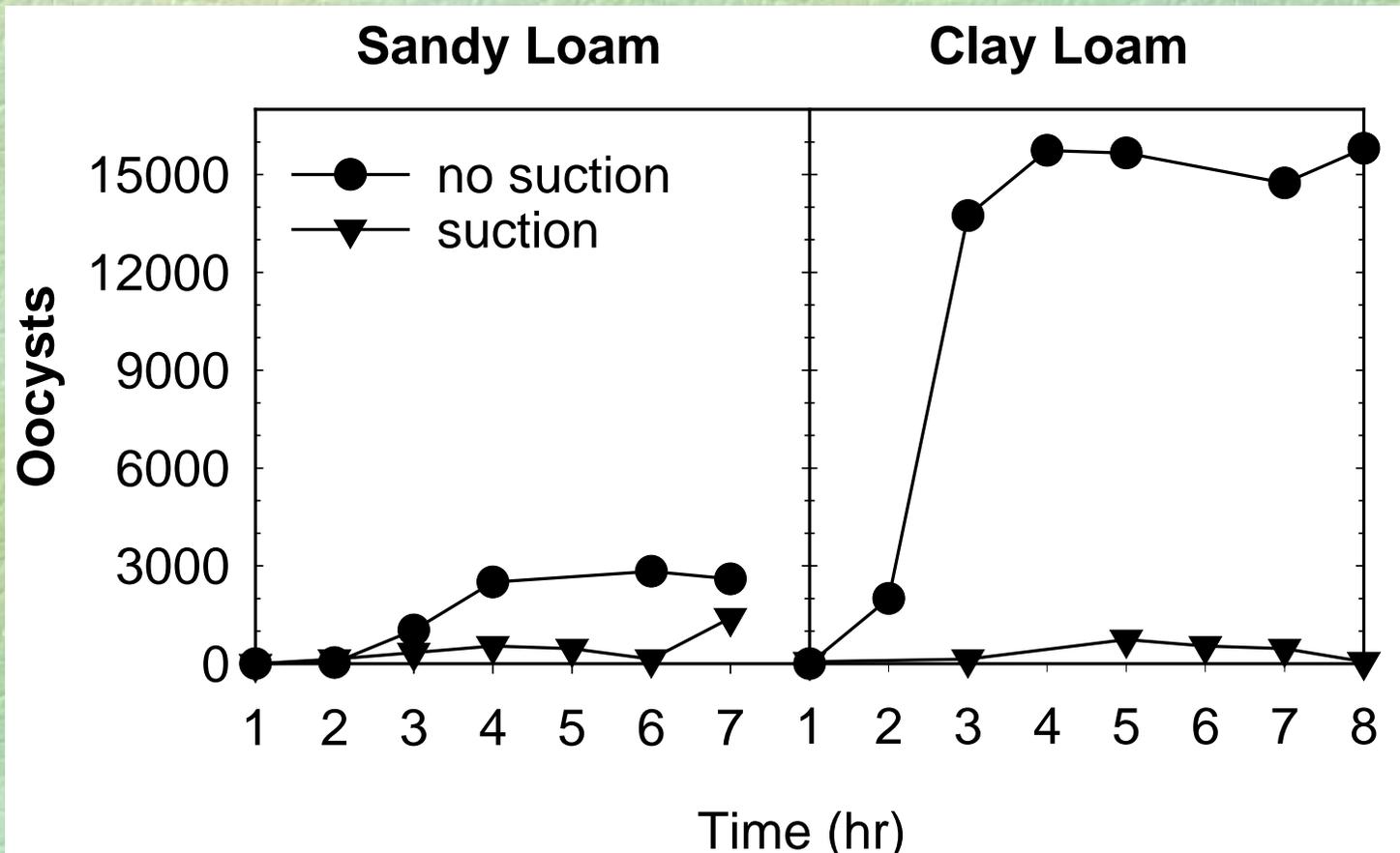
$\mu$  = the combined first-order removal rate constant

$c_m$  = the concentration of oocyst released from manure

# Parameter estimation

- 1. Values of  $\theta$  and  $\nu$  were measured.**
- 2. Values of  $\lambda$  were computed from the percentage of oocysts left on the soil surface.**
- 3. The value of  $D$  was computed as  $D=l\nu$  with  $l=1$  cm and  $l=0.5$  cm in saturated and unsaturated samples, respectively.**
- 4. The analytical solution (van Genuchten, 1985) was fitted to data on profile oocyst distributions and cumulative amount of oocysts in leachate to estimate  $R$  and  $\mu$**

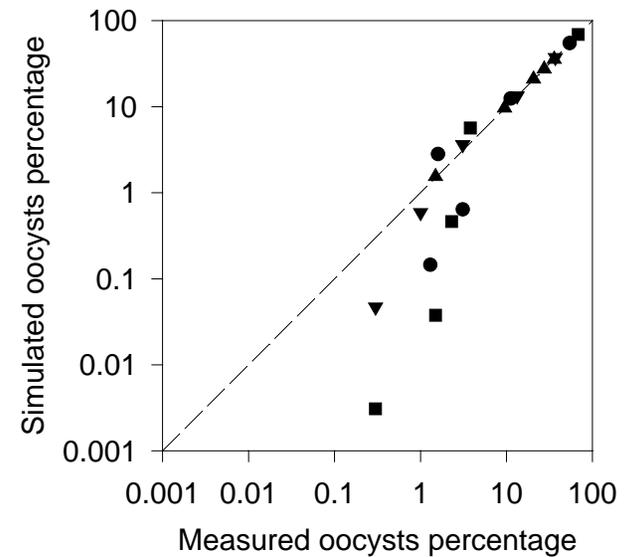
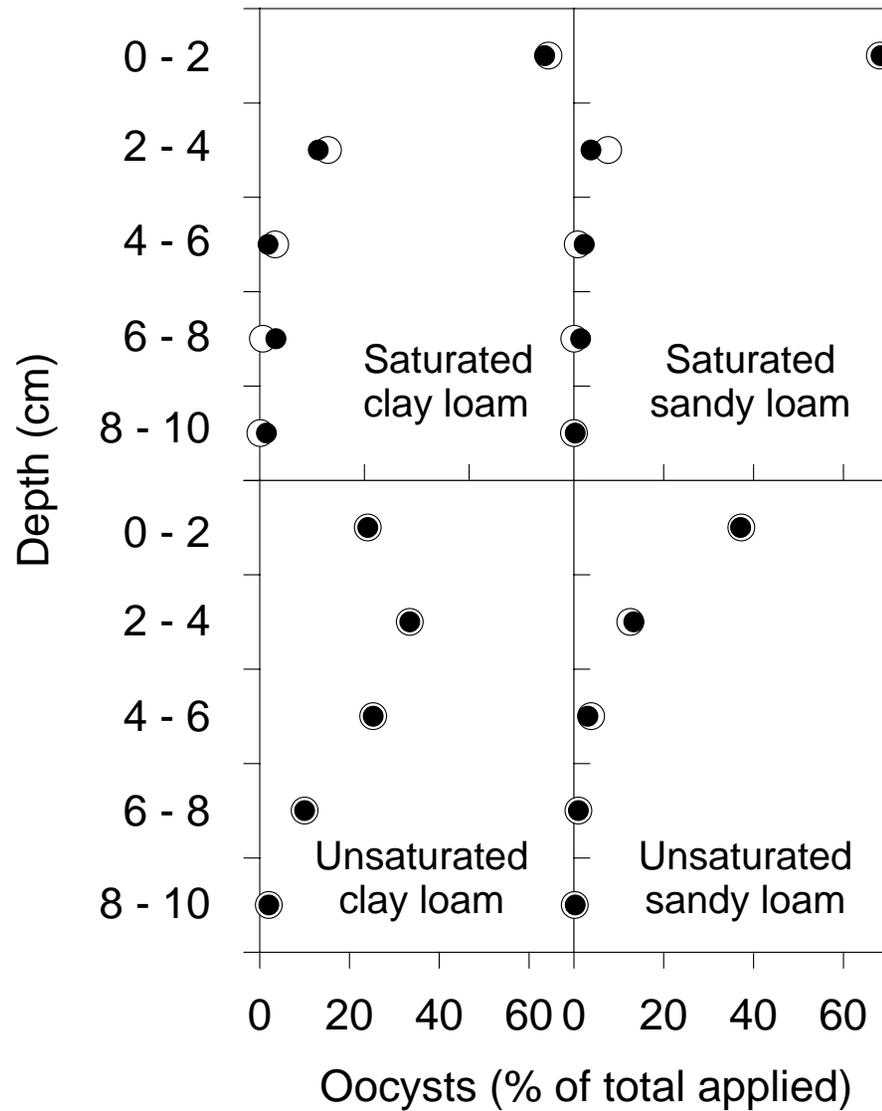
# Oocyst leaching



## Cumulative leaching (%)

	Clay loam	Sandy loam
Saturated	1.3	0.4
Unsaturated	<0.1	<0.1

# Measured (●) and simulated (○) oocyst profile distributions



# Transport parameters

Parameter	Saturated columns		Unsaturated columns	
	Clay loam	Sandy loam	Clay loam	Sandy loam
$\nu$ (cm h <sup>-1</sup> )	2.7	2.6	3.3	1.5
$D$ (cm <sup>2</sup> h <sup>-1</sup> )	2.7	2.6	1.7	0.7
$\lambda$ (h <sup>-1</sup> )	0.212	0.503	0.240	0.134
$\mu^{\dagger}$ (h <sup>-1</sup> )	2.77 - 4.13	5.28 - 9.96	0.00 - 0.16	0.62 - 0.81
$R^{\dagger}$	0.00 - 0.97	0.00 - 1.56	4.51 - 4.61	2.44 - 3.82

<sup>†</sup>Confidence intervals of the optimized values at the 5% significance level

Release of manure colloids:  $\lambda=0.12-0.18$  h<sup>-1</sup> (Bradford and Schijven, 2002),  
 $\lambda=0.32$  h<sup>-1</sup> (Shelton et al., 2002).

Oocyst removal rates: 2 to 30 h<sup>-1</sup> at  $\nu=6 - 60$  cm h<sup>-1</sup> (Harter et al., 2000, sands),  
 1500 to 3000 h<sup>-1</sup> at  $\nu= 40,000$  cm h<sup>-1</sup> (Brush et al., 1999, glass beads)

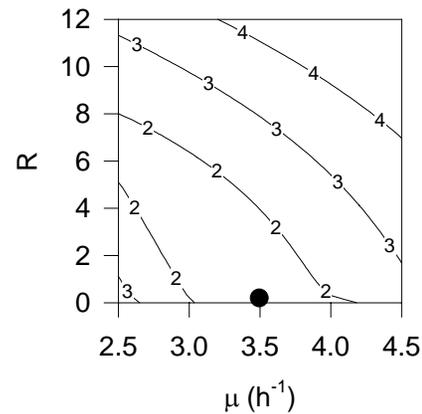
Removal rates and retardation coefficients are markedly different in saturated and unsaturated columns.

$K_d = 1.4 -7.7$  from column data,  $K_d$  between 100 and 700 in batch experiments

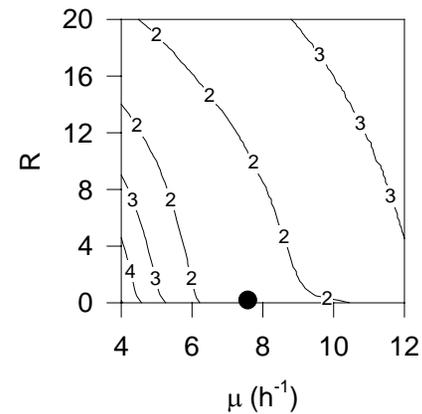
# Challenge for parameter estimation

The same overall model error can be achieved with various combinations of  $R$  and  $\mu$ .

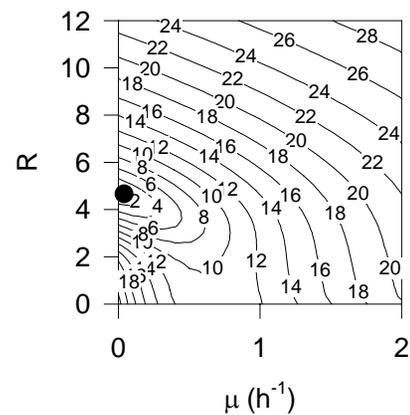
**Saturated clay loam**



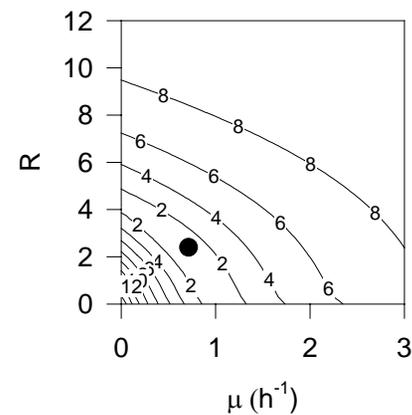
**Saturated sandy loam**



**Unsaturated clay loam**



**Unsaturated sandy loam**



# Conclusions

**Presence of manure markedly affects attachment of *C. parvum* oocysts to soil.**

**Soil texture is a substantial factor of oocyst attachment in batch experiments; soil structure may be more important for retention during transport**

**The breakthrough of oocysts released with manure particulates was higher in saturated than in unsaturated soil**

**The convective-dispersion model simulated the profile distributions of oocysts reasonably well, but failed to simulate breakthrough of small amounts of oocysts occurring probably due to a preferential flow**

**Values of retardation coefficient were most probably less than unity in saturated columns and greater than unity in unsaturated columns.**

**The removal rates were much higher in saturated columns as compared with unsaturated columns.**

**Oocyst adsorption data from batch experiments may be inapplicable in transport models that employ the adsorption equilibrium hypothesis.**